Claims

1. A recombinant nucleotide sequence which codes upon expression for at least a part of a bifunctional hybrid active-site serine ß-lactamase protein, wherein the ß-lactamase protein is bearing at least one heterologous sequence, **characterized** in that the hybrid protein is having two functions, the first function is associated with the ß-lactamase portion and the second function is associated with the heterologous sequence having a biological function which is different from the first function.

2. The recombinant nucleotide sequence according to claim 1, wherein the ß-lactamase protein is having conserved amino acid elements 1, 2 and 3, wherein element 1 is having the amino acid sequence SXXK, element 2 is having the amino acid sequence SDN in class A proteins, YXN in class C proteins, SX[V or T or N] in class D proteins, wherein the elements of classes A, C and D correspond to each other, and element 3 is having the amino acid sequence K[T or S]G, **characterized** in that the ß-lactamase protein is bearing at least one heterologous sequence between element 2 and element 3.

3. The recombinant nucleotide sequence according to claim 1 or 2, **characterized** in that the ß-lactamase protein is bearing at least one heterologous sequence in a region located between two neighboring alpha helices of the ß-lactamase sequence, wherein the region is forming a juncture between the alpha helices of active-site serine ß-lactamases, wherein said alpha helices correspond to the last two alpha helices before the alpha/beta domain.

4. The recombinant nucleotide sequence according to any of claims 1 to 3, **characterized** in that the ß-lactamase protein is bearing at least one heterologous sequence in a region located between two neighboring alpha helices of the ß-lactamase sequence, wherein the region is selected from:
a) the region forming a juncture between alpha helix 8 and alpha helix 9 of TEM-1 ß-lactamase;
b) the region forming a juncture between the alpha helices which are homologous to alpha helix 8 and alpha helix 9 of TEM-1 ß-lactamase.

5. The recombinant nucleotide sequence according to any of claims 1 to 4, **characterized** in that the ß-lactamase moiety is selected from the goup:
a) class A ß-lactamase,
b) class C ß-lactamase,
c) class D ß-lactamase,
d) a recombinant sequence of one or more of a) to c).

6. The recombinant nucleotide sequence according to any of claims 1 to 5, **characterized** in that the ß-lactamase moiety is derived from class A ß-lactamase, wherein ß-lactamase class A protein is bearing the heterologous sequence in the region forming a juncture between alpha helix 8 and alpha helix 9.

7. The recombinant nucleotide sequence according to claim 6, **characterized** in that the region forming a juncture between alpha helix 8 and alpha helix 9 is selected from the group:
a) the amino acid sequence Thr195 to Leu199 of the TEM-1 ß-lactamase;
b) the amino acid sequence corresponding to the amino acid sequence Thr195 to Leu199 in TEM-1 ß-lactamase.

8. The recombinant nucleotide sequence according to any of claims 1 to 5, **characterized** in that the ß-lactamase moiety is derived from class C ß-lactamase, wherein ß-lactamase class C protein is bearing the heterologous

sequence in the region forming a juncture between alpha helices, which correspond to alpha helix 8 and alpha helix 9 in TEM-1 ß-lactamase.

9. The recombinant nucleotide sequence according to claim 8, **characterized** in that the region forming a juncture is selected from the group:
a) the amino acid sequence K239 to E245 of the AmpC ß-lactamase;
b) the amino acid sequence corresponding to the amino acid sequence K239 to E245 of the AmpC ß-lactamase.

10. The recombinant nucleotide sequence according to any of claims 1 to 5, **characterized** in that the ß-lactamase moiety is derived from class D ß-lactamase, wherein ß-lactamase class D protein is bearing the heterologous sequence in the region forming a juncture between alpha helices, which correspond to alpha helix 8 and alpha helix 9 in TEM-1 ß-lactamase.

11. The recombinant nucleotide sequence according to claim 10, **characterized** in that the region forming a juncture is selected from the group:
a) the amino acid sequence N510 to F514 of the BlaR-CTD ß-lactamase;
b) the amino acid sequence corresponding to the amino acid sequence N510 to F514 of the BlaR-CTD ß-lactamase.

12. A recombinant nucleotide sequence which codes upon expression for at least a part of a bifunctional hybrid ß-lactamase class A protein, **characterized** in that the ß-lactamase class A protein is bearing at least one heterologous sequence in a region located between two neighboring alpha helices of the ß-lactamase sequence, wherein the region is selected from:
a) the region forming a juncture between alpha helix 8 and alpha helix 9 of the TEM-1 ß-lactamase;
b) the region forming a juncture between the alpha helices of homologous ß-lactamases class A, said alpha helices corresponding to the alpha helix 8 and alpha helix 9 of the TEM-1 ß-lactamase.

14. The recombinant nucleotide sequence according to any one of claims 1 to 13, wherein the hybrid protein is having two functions, the first function is associated with the ß-lactamase portion and is selected from

c) hydrolyzing ß-lactams (ß-lactamase activity);

d) binding covalently and in a stable manner to substances selected from the group ß-lactams, derivatives of ß-lactams, inhibitors of ß-lactams;

wherein the second function is associated with the heterologous sequence having a biological function which is different from the first function.

15. The recombinant nucleotide sequence according to any one of claims 1 to 14, wherein the three-dimensional structure of the ß-lactamase portion of the hybrid ß-lactamase is homologous to the three-dimensional structure of the TEM-1 ß-lactamase.

16. The recombinant nucleotide sequence according to any one of claims 1 to 15, wherein the heterologous sequence has a length of 11 or more amino acid residues.

17. The recombinant nucleotide sequence according to any one of claims 1 to 15, wherein the heterologous sequence has a length of 18 or more amino acid residues.

18. The recombinant nucleotide sequence according to any one of claims 1 to 15, wherein the heterologous sequence has a length of 25 or more amino acid residues.

19. The recombinant nucleotide sequence according to any one of claims 1 to 15, wherein the heterologous sequence has a length of 50 or more amino acid residues.

20. The recombinant nucleotide sequence according to any one of claims 1 to 15, wherein the heterologous sequence has a length of 100 or more amino acid residues.

21. The recombinant nucleotide sequence according to any one of claims 1 to 20, wherein the nucleotide sequence coding for the ß-lactamase sequence is selected from:

a) nucleotide sequence coding for the ß-lactamase TEM-1 (SEQ ID NO: 1)
b) nucleotide sequence coding for the ß-lactamase BlaP (SEQ ID NO: 2);
c) nucleotide sequence coding for the ß-lactamase BlaL (SEQ ID NO: 3);
d) nucleotide sequence coding for the ß-lactamase AmpC (SEQ ID NO: 39);
e) nucleotide sequence coding for the ß-lactamase BlaR-CTD (SEQ ID NO: 41);
f) a recombinant sequence of one or more of a) to e);
g) nucleotide sequences which hybridise under stringent conditions to the nucleotide sequences of any one of a) to f) or fragments thereof.

22. The recombinant nucleotide sequence according to any one of claims 1 to 21, wherein the heterologous sequence is related to a function selected from: being an epitope, being a specific binding partner for antibodies, being specifically recognized and bound by antibodies, having a binding affinity to earth alkali and metal ions, having enzymatic activity, being a toxin (STa heat-stable enterotoxin of E. coli), bearing a glycosylation site, bearing a glycosylated peptide, being a specific binding partner for any polypeptide or any ligand, having a binding affinity to dsDNA and ssDNA or RNA (having a binding affinity to nucleotide and polynucleotide).

23. The recombinant nucleotide sequence according to any one of claims 1 to 22, wherein the heterologous sequence is selected from the group: STa (heat stable enterotoxin of *Escherichia coli*, SEQ ID NO: 21), protein A of *Staphylococcus aureus,* (SEQ ID NO: 23 and 25), protein G of *Streptococcus pyogenes,* (SEQ ID NO: 27 and 29), a linear antigenic

determinant of the hemagglutinin of the Influenca virus (SEQ ID NO: 31), a fragment of human phospholipase – type II ($hPLA_2$) (SEQ ID NO: 33), LPS binding amino acid sequence (SEQ ID NO: 35), and nucleotide sequences which hybridise under stringent conditions to said nucleotide sequences or fragments thereof.

24. A recombinant polypeptide which is encoded by the recombinant nucleotide sequence according to any one of claims 1 to 23.

25. A recombinant polypeptide comprising at least a part of a bifunctional hybrid active-site serine ß-lactamase protein, wherein the ß-lactamase protein is bearing at least one heterologous sequence, **characterized** in that the hybrid protein is having two functions, the first function is associated with the ß-lactamase portion and the second function is associated with the heterologous sequence having a biological function which is different from the first function.

26. The recombinant polypeptide according to claim 25, wherein the ß-lactamase protein is having conserved amino acid elements 1, 2 and 3, wherein element 1 is having the amino acid sequence SXXK, element 2 is having the amino acid sequence SDN in class A proteins, YXN in class C proteins, SX[V or T or N] in class D proteins, wherein the elements of classes A, C and D correspond to each other, and element 3 is having the amino acid sequence K[T or S]G, **characterized** in that the ß-lactamase protein is bearing at least one heterologous sequence between element 2 and element 3.

27. The recombinant polypeptide according to to claim 25 or 26, **characterized** in that the ß-lactamase protein is bearing at least one heterologous sequence in a region located between two neighboring alpha helices of the ß-lactamase sequence, wherein the region is forming a juncture between the alpha helices of active-site serine ß-lactamases, wherein said alpha

helices correspond to the last two alpha helices before the alpha/beta domain.

28. The recombinant polypeptide according to any of claims 25 to 27, **characterized** in that the ß-lactamase protein is bearing at least one heterologous sequence in a region located between two neighboring alpha helices of the ß-lactamase sequence, wherein the region is selected from:
a) the region forming a juncture between alpha helix 8 and alpha helix 9 of TEM-1 ß-lactamase;
b) the region forming a juncture between the alpha helices which are homologous to alpha helix 8 and alpha helix 9 of TEM-1 ß-lactamase.

29. The recombinant polypeptide according to any of claims 25 to 28, **characterized** in that the ß-lactamase moiety is selected from the goup:
a) class A ß-lactamase,
b) class C ß-lactamase,
c) class D ß-lactamase,
d) a recombinant sequence of one or more of a) to c).

30. The recombinant polypeptide according to any of claims 25 to 29, **characterized** in that the ß-lactamase moiety is derived from class A ß-lactamase, wherein ß-lactamase class A protein is bearing the heterologous sequence in the region forming a juncture between alpha helix 8 and alpha helix 9.

31. The recombinant polypeptide according to any of claims 25 to 29, **characterized** in that the ß-lactamase moiety is derived from class C ß-lactamase, wherein ß-lactamase class C protein is bearing the heterologous sequence in the region forming a juncture between alpha helices, which correspond to alpha helix 8 and alpha helix 9 in TEM-1 ß-lactamase.

32. The recombinant polypeptide according to any of claims 25 to 29, **characterized** in that the ß-lactamase moiety is derived from class D ß-

lactamase, wherein ß-lactamase class D protein is bearing the heterologous sequence in the region forming a juncture between alpha helices, which correspond to alpha helix 8 and alpha helix 9 in TEM-1 ß-lactamase.

33. A recombinant polypeptide comprising at least a part of a bifunctional hybrid ß-lactamase class A protein, **characterized** in that the ß-lactamase class A protein is bearing at least one heterologous sequence in a region located between two neighboring alpha helices of the ß-lactamase sequence, wherein the region is selected from:
a) the region forming a juncture between alpha helix 8 and alpha helix 9 of the TEM-1 ß-lactamase;
b) the region forming a juncture between the alpha helices of homologous ß-lactamases class A, said alpha helices corresponding to the alpha helix 8 and alpha helix 9 of the TEM-1 ß-lactamase.

34. The recombinant polypeptide according to any of claims 25 to 33, wherein the hybrid ß-lactamase is possessing an activity selected from
a) hydrolysing ß-lactams;
b) binding covalently and in a stable manner to derivatives of ß-lactams and inhibitors.

35. The recombinant polypeptide according to any one of claims 25 to 34, wherein the three-dimensional structure of the ß-lactamase portion of the hybrid ß-lactamase is homologous to the three-dimensional structure of the TEM-1 ß-lactamase.

36. The recombinant polypeptide according to any one of claims 13 to 16, wherein the ß-lactamase sequence is selected from:
a) ß-lactamase TEM-1 (SEQ ID NO: 4)

b) ß-lactamase BlaP (SEQ ID NO: 5);

c) ß-lactamase BlaL (SEQ ID NO: 6);

d) ß-lactamase AmpC (SEQ ID NO: 38);

e) ß-lactamase BlaR-CTD (SEQ ID NO: 40).

37. The recombinant polypeptide according to any one of claims 25 to 26, wherein the heterologous sequence is related to a function selected from: being an epitope, being a specific binding partner for antibodies, being specifically recognized and bound by antibodies, having a binding affinity to earth alkali ions and metal ions, having enzymatic activity, being a toxin (STa heat-stable enterotoxin of E. coli), bearing a glycosylation site, bearing a glycosylated peptide, being a specific binding partner for any polypeptide or any small ligand, having a binding affinity to dsDNA and ssDNA or RNA (having a binding affinity to nucleotide and polynucleotide).

38. The recombinant polypeptide according to any one of claims 25 to 37, wherein the heterologous sequence is selected from the group: STa (heat stable enterotoxin of Escherichia coli) (SEQ ID NO: 22), protein A of *Staphylococcus aureus* (SEQ ID NO: 24 and 26), protein G of *Streptococcus pyogenes* (SEQ ID NO: 28 and 30), a linear antigenic determinant of the hemagglutinin of the Influenca virus (SEQ ID NO: 32), a fragment of human phospholipase – type II ($hPLA_2$) (SEQ ID NO: 34), LPS binding amino acid sequence (SEQ ID NO: 36).

39. Use of the recombinant nucleotide sequence of claims 1 to 24 or the recombinant polypeptide of any one of claims 25 to 38 for vaccination.

40. Use of the recombinant nucleotide sequence of claims 1 to 24 or the recombinant polypeptide of any one of claims 25 to 38 for raising antibodies against the heterologous sequence.

41. Use of the recombinant nucleotide sequence of claims 1 to 24 or the recombinant polypeptide of any one of claims 25 to 38 for epitope mapping.

42. Use of the recombinant nucleotide sequence of claims 1 to 24 or the recombinant polypeptide of any one of claims 25 to 38 for affinity chromatography.

43. Use of the recombinant nucleotide sequence of claims 1 to 24 or the recombinant polypeptide of any one of claims 25 to 38 for the concentration and/or purification of antibodies directed against the heterologous sequence.

44. Use of the recombinant nucleotide sequence of claims 1 to 24 or the recombinant polypeptide of any one of claims 25 to 38 for the qualitative and/or quantitative detection of molecules binding to the heterologous sequence.

45. Use of according to claim 44, wherein the molecules binding to the heterologous sequence are antibodies or antibody fragments, polypeptides, dsDNA, ssDNA, RNA or small ligands.

46. Pharmaceutical compositions comprising a recombinant polypeptide of any one of claims 25 to 38.

47. Use of a recombinant polypeptide of any one of claims 25 to 38 for the manufacture of a medicament for the preventive and/or therapeutic treatment of diseases selected from the group cancer, viral diseases and bacterial diseases (or infection diseases), autoimmune diseases and allergy.

48. The use of a recombinant polypeptide of any one of claims 25 to 38 for the development of a medicament

49. A method for screening a compound for treatment, prevention and/or diagnosis of a disease which comprises the step of detecting interaction between the homologous sequence of the hybrid ß-lactamase according to claims 25 to 38 and a protein or polypeptide which binds to the homologous sequence in presence of a compound to be tested.

50. The method according to claim 49, wherein the compound tested is selected as a candidate of an effective medicament when the compound has an effect on the interaction between the homologous sequence and the polypeptide which binds to the homologous sequence.

51. The method according to claims 49 or 50, which comprises the steps of:

a) subjecting the recombinant polypeptide of any one of claims 25 to 38 and a polypeptide which binds to the homologous sequence to interaction with each other in the presence of the compound to be tested;

b) subjecting the recombinant polypeptide of any one of claims 25 to 38 and a polypeptide which binds to the homologous sequence to interaction with each other in the absence of the compound to be tested;

c) detecting the interactions in the steps a) and b), and

d) comparing the interactions in the steps a) and b) to chose the compound having an effect on the interaction as a candidate of an effective medicament.

52. A biologic sensor comprising a recombinant polypeptide of any one of claims 25 to 38.

53. The biologic sensor according to claim 33, wherein the biologic sensor is comprising a carrier bearing a recombinant polypeptide of any one of claims 25 to 38.